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Notes and Observations on the Red Streak

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NOTES AND OBSERVATIONS ON THE RED STREAK ASSOCIATED WITH OUEENSLAND TOP ROT DISEASE.

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Introduction.

The experiments and observations outlined below were carried out with a view to inquiring into the nature of certain bright Red Streaks which appear in cane to inquiring into the nature of certain bright Red Streams which appear in case leaves, especially Badila (N.G. 15), at about this time of the year. The whole of the inquiry was carried out during the very short period of three weeks—15th December, 1925, to 6th January, 1926—so that the work had to be done in a more or less hurried fashion, and it is only because it is probable that a considerable period of time must elapse before the present writer can again take up the work that these preliminary notes are being published.



PLATE 119 (Fig. 1).

During the course of this paper the words Red Streak, written with capital letters, refer definitely to the Red Streaks under investigation, and only to such, while red streak written without capitals is to be considered as a collective term referring to any of the many different types of red streaks which accompany cane ailments.

The writer wishes to thank the Rev. N. Michael, who, by giving the use of a room, stove, cupboards, &c., greatly facilitated the execution of the microscope work and culture work necessitated by this inquiry, and Mr. D. S. North, pathologist to the Colonial Sugar Refining Company, Limited, for valuable suggestions.

To Mr. H. Ritchie, of Kilrie Farm, and to his neighbour, Mr. J. Soper, junr., many thanks are also due for permission to carry out the inoculation experiments in

Description of Red Streak Infection.

The first visible indication of Red Streak infection in a cane top is the presence at the base of one or more leaf blades, usually on the half unfurled or first unfurled leaf, of a narrow, dark watery green, longitudinal streak about 1 in. to $1\frac{1}{2}$ in. in length and $\frac{1}{16}$ in. in width. This streak grows very quickly, and as it grows alters in

colour, gradually changing from watery green to watery brown and then to bright blood red. Fresh streaks are meanwhile formed on the same and younger leaves, and unless some climatic or other factor which is not yet understood intervenes the infection apparently progresses until all of the younger leaves are blazoned with brilliant red lines, which will sometimes coalesce if two or more streaks be close together, forming bands often ½ in. or more in width and 2 ft. or 3 ft. in length. (See Fig. 1.) On the other hand, the infection will sometimes cease to be active after one thin streak, perhaps no more than 1 in. in length, has been formed.

Red Streaks are able to form in any portion of the leaf (see Fig. 2), though it is the general rule for them to take their source at or near the base of the blade; they are often found on the under side of the mid-rib, and these are the ones that usually attain the greatest length.

When the Red Streaks become old the tissue which originally formed them will sometimes die and wither, and assume a chocolate-brown colour.

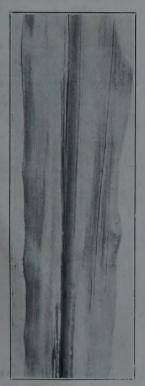




PLATE 120 (Fig. 2 and 3).

A very noticeable feature of the most active Red Streaks is that they are almost invariably splashed here and there along their whole length by brown or white stains which appear to be the result of the drying of some exudation formed during their growth, though no wounds of any kind are visible at these places, unless the leaf tissue has broken down completely, as sometimes does happen.

Observations on Growth of Red Streak.

With a view to observing the growth of Red Streaks three stems were selected, marked, and examined from time to time. Observations were commenced at 12 noon on 19th December, when stem

A had one sharp narrow Red Streak 6 in. long, and commencing about 11 in. from leaf-sheath juncture on third fully unfurled leaf.

- B had one 2½ in. streak of a watery green colour tinged here and there with pink on half unfurled leaf.
 - C, which was part of a badly Red Streak infected stool, one stalk having already been killed by Top Rot, was marked by one streak 9 in. long, bright red, and starting from near leaf-sheath juncture.

21st December-

- A Streak unaltered.
- B Streak was now 12 in. long, bright red at base, and very dark watery green to watery brown spotted with red on upper half, and had a vague yellowish tip and background.
- C Original streak unaltered. Four fresh streaks about 2 in. in length bad formed immediately adjacent to the older streak and with their bases at the leaf-sheath juncture; their colour varied from bright red to watery brown.

23rd December-

- A No alteration.
- B Original streak now 18 in. long and all dark red, with withered portions in upper part. Fresh watery green to brown streak 4 in. long now present immediately to right of above. The second leaf above the originally infected leaf—i.e., what was now the first fully unfurled leaf—now showed a patch about 1 in. long by \(\frac{1}{2} \) in. wide about \(\frac{1}{2} \) in. above the leaf-sheath juncture. Central portion of this patch was of a light brown colour, while edges were a light pink. This patch apparently represented a severe local infection
- C Streaks all dark red, otherwise no alteration.

26th December -- -

- A No alteration.
- B A fresh young streak about 2 in, long was now forming on leaf situated between two leaves already infected, while the patch referred to above had become part of a wide vigorous looking streak 4 in, in length.
- C No alteration.

4th January-

- A No alteration.
- B Position of infected leaves was now 3rd, 4th, and 5th, fully unfurled. Streaks on 5th (original infected leaf) had coalesced to form a large dark Red Streak 18 in. long, with dead central tissue in places. Streak on 4th was now about 8 inches long and all dark red. Third leaf was still showing signs of activity, as two watery brown streaks each about 2 in. long were forming at base of leaf. Most of patch noticed on this leaf on 23rd December was now quite dead and largely disrupted, while the streak emanating therefrom was now some 10 in. long and marked every here and there by a brown or white "exudation stain."
- C No alteration.

Distribution of Red Streaks in a Field.

Some counts were taken in the field where the above observations were made of three rows, each ten rows apart, in the worst infected end of the field. Thus—

Row	7. Total Number Stools.	Percentage of Diseased Stools.	Total Number Stems in Diseased Stools.	Percentage of Diseased Stems.
1	191	30.0	270	38
2	287	19.5	. 302	43
3	290	5.5	89	21

Further counts taken a week later showed but very slight increase in the incidence of the Red Streaks, though, judging by appearances, the number of Red Streaks had increased very much.

Though single infected stools were met with here and there, it was very noticeable that most of the diseased stools in each row were confined to four or five well-defined centres of infection.

Most of the infected stools were situated in the north-eastern corner of this field, where the cane was showing the best and most vigorous growth.

However, Red Streaks are not necessarily always confined to the most vigorous cane, or even to young plant cane. The writer has seen a field of young plant Badila at Stratford, near Cairns, very seriously infected with Red Streaks, though the cane at Stration, hear Carries, very seriously intected with Red Streaks, though the cane was very backward indeed, and growing on badly-drained, sour-smelling land. On the Burdekin River Red Streaks were observed on this visit on the leaves of fully-matured Badila, while on the Herbert River late in November Red Streaks were seen on young ratoon shoots 18 in. high of Badila and Q. 813. Vigorous young plant Badila, however, is the cane usually attacked.

Relationship of Red Streaks to Top Rot Disease.

A field which is well infected with Red Streaks usually gives off a strong smell of decaying fruit, and upon close examination it is seen that every here and there a stalk whose top is literally covered in Red Streaks has a dead "heart," which may be easily pulled out (see Fig. 1), its lower end being quite rotted, and which has an offensive



PLATE 121 (Fig. 4).

odour. On the other hand, some tops are found whose hearts are dead but whose beaves are not marked by Red Streaks; however, when the heart is pulled out it is found that some of the very young leaves, now dead, are marked, at a region usually about their middle, by a peculiar streaky discoloured area distinctive in the very irregularity of its form; such areas do not generally cover more than about 3 square inches, usually much less. Both of these conditions constitute what is known as Top

During this inquiry it was observed that fully 90 per cent, of the stalks whose hearts had been killed by Top Rot had one or more leaves marked by one or more Red Streaks.

Questioned on the matter of relationship of Red Streaks to Top Rot, some twenty farmers were adamant in the expressed opinion that Red Streaks were the early stage of Top Rot, and that fields which later suffered heavily from Top Rot always showed a heavy infestation of Red Streaks before the real damage was done, though the fact that Red Streaks were present did not always signify that Top Rot would be severe later on—i.e., that cane often appeared to be able to recover from a severe visitation of Red Streaks and show but very little loss from Top Rot.

H. Tryon (1), in his paper on Top Rot Disease, refers to Red Streaks in four places (pp. 8, 22, and 41), and in one place at least (p. 22), where one grower is quoted as having remarked:—''Yes, I was amongst this cane with Red Streaks in its leaves five weeks since, and was then able by the odour that it emitted to recognise the presence of Top Rot.'' It seems that the Red Streaks described above are being referred to.

More recently the writer (2) has referred to Red Streaks as being the early stage of Top Rot, basing his opinions on field observations.

That there is some intimate relationship between Red Streaks and Top Rot is obvious since the former seem to invariably precede and accompany the latter in the field, though not necessarily in the stalk.

Similar Diseases in Other Countries.

In Hawaii a disease occurs which is known as Bacterial Red Stripe Disease, and bears many characteristics similar to those of Top Rot as it is known in Queensland. II, Atherton Lee and W. C. Jennings (3) state that this disease "is easily identified by the long, narrow, dark red, longitudinal streaks on the cane leaves. These streaks usually start midway between the tip of the leaf and its juncture with the leaf sheath, at the point where the lend in the leaves of Tip Canes takes place. The first indication of the disease is a watery darkened streak, not yet red, but still green, which spreads longitudinally up and down the leaf. This watery, dark green streak gradually becomes bright red in colour."

Polvillo is a bacterial disease which occurs in Tucuman, Argentine, and has been carefully studied by G. L. Fawcett (4). The description of this disease very much resembles that of Top Rot. As in the case of the latter disease red stripes and dead hearts are produced. Plant came chiefly is attacked.

D. S. North (5) points out the similarity between Top Rot, Red Stripe, and Polvillo, in the following words:—''Conspicuous red streaks are the most characteristic symptoms with all three diseases. Those of Red Stripe usually start about the middle of the leaf blade, and are long, whereas those of Top Rot are shorter, and are usually confined to the last of the leaf blade, although in other respects much the same. Reddening of the leaf-sheath occurs regularly with both Polvillo (the Argentine disease) and Top Rot, but only in severe cases with Red Stripe. All three cause a Top Rot in severe cases. Cuttings from diseased stalks do not usually produce diseased plants. When Top Rot occurs side shoots of healthy appearance may sprout from below the rotten portion and grow without further sign of the disease.'' Referring to Red Stripe disease the same author goes on to say that—'In these and all other respects the resemblance is so striking that we may now suspect our Top Rot to be due to a similar cause, vir., bacteria, which are probably disseminated by the wind and flying insects, and which need rainy weather in order to infect the leaves. Frequently the disease is confined to the leaves thus affected, and disappears when these leaves wither, unless younger leaves become similarly infected in the meantime. Even in severe cases where Top Rot results from the invasion of the apical bud and top generally, the disease runs a rapid course and disappears with a change to weather conditions unfavourable to it, leaving the top dead but the rest of the stalk free from infection.''

It was especial consideration of the paper just quoted which led the writer to investigate the matter in the field on this occasion...

Before going further the writer would like to mention that in his experience the leaf-sheaths of Red Streak infected cane have not become reddened under field conditions except in fairly severe cases.

Were Red Streaks and Top Rot proved to be infectious bacterial diseases, a moment's consideration would show us what ideal conditions obtain at the season of Red Streak appearance for the transmission and incubation of the responsible organism. The water of transpiration always present on cane tops during the season when the disease is prevalent provides a suitable channel of infection and incubation if such are needed, and, again, the under side of the base of the blades of young cane leaves is the favourite feeding ground of certain sap-sucking Homoptera, such as Perkinsiella saccharicida, which also makes large punctures in the mid-rib when laying its eggs.

Bacteria Found in Red Streaks Associated with Queensland Top Rot.

Late in November last the writer examined Red Streaks from young ration Badila and Q. 813 at Macnade, Herbert River, and found many active bacteria in all of them, but especially in those streaks which had reached about the half-way stage in the transition from watery green to red.

Red Streaks examined by the writer at Ayr on 16th December were similarly found to contain many active bacteria.

Reproduction of Red Streaks by Artificial Inoculation.

SERIES I.

16th December.—A typical young streak, i.e., one transforming from watery green to red, was suitably cleaned and a portion crushed up in sterilised water. The suspension so formed was used for inoculating sterilised potato slices. In twenty-four to thirty hours a thick, viscid, creamy yellow growth had formed. Lack of bacteriological equipment prevented the preparation of cultures aiming at the isolation of the organism in pure culture.



PLATE 122 (Fig. 5).

19th December,—After three days' growth on a potato slice a suspension of the bacteria was made in sterilised water and used for inoculating the leaves of fifteen stalks of Badila (N.G. 15) in a field of vigorously growing eight months old irrigated plant cane showing no definite Red Streaks. The suspension contained actively motile bacteria of similar appearance to those seen in Red Streaks previously.

The methods of inoculation included inoculation with and without punctures and with a protective covering, inoculations a few inches up one or two leaves of individual stalks, and inoculation at the base of the heart. Punctures were made with a sterilised pricker, and covering was effected by placing a little damp cotton wool on each side of inoculated region and wrapping the two or more inoculated leaves and those adjoining with thin waxed paper.

Development of Red Streaks in Inoculated Leaves.

Within seven days 100 per cent, of punctured leaves and 75 per cent, i.e., 3, of stalks inoculated without wounding showed positive infection of Red Streaks.

All wrappings were removed on the second day, when it was found that in most cases the cotton wool was still damp. A short, watery green streak had now formed near the base of a half-unfurled leaf on one stalk, while in most other cases it was observed that the punctures, where visible, were surrounded by a light red margin.

On the fourth day nine stems, including one which had been inoculated by simply placing a drop of suspension on the under side of the half-unfurled leaf and on the upper side of the leaf immediately below it, and two in which inoculation had consisted of pouring a little suspension on to the base of the heart, showed one or more watery green to light yellow streaks on their leaves. Even where the needle had been used it was observed that not all of these streaks arose from punctures, as in some cases streaks had developed in regions of the leaf unwounded throughout their length. Many of the older streaks, i.e., those streaks which were already approaching a red colour, were marked by one or more of the "exudation stains" described earlier. Five other stems were now developing watery green streaks about punctures.

On the fifth day three stems showed positive infection with Red Streaks. In all cases the infection was developing rapidly. Fresh "exudation stains" were seen to have formed.

On the sixteenth day it was found that the infection had been so vigorous that many leaves bore Red Streaks 1 ft, to 3 ft, in length, and in some cases fresh watery green streaks were still forming on the green leaves of heart. In few cases had the infection travelled below the leaf-sheath junctures.

19th December.—The leaves of eight stalks near those treated in Series I. were similarly inoculated as controls, using as inoculum a brick-red bacterial colony which had developed on a control potato slice through contamination.

Red Streaks were sparsely produced on five of these stalks, however, being possibly attributable to a carry-over of bacteria from the earlier inoculations in spite of the aseptic precautions taken.

SERIES III.

This series of inoculations was carried out at the least infected end of the field where the infection counts mentioned earlier were taken in well-watered, vigorously growing, eight months old Badila. Very few stalks showed Red Streaks in the vicinity of those inoculated.

21st December.—Ten stalks were inoculated direct from infected leaves taken from the field itself by drawing a needle through a vigorous watery brown streak and then scratching the half-unfurled leaf and that immediately below it at about 4 in. from the leaf-sheath junctures.

Twenty stalks were inoculated as in Series I., using a pricker and a suspension made from a five days old culture.

Of these thirty stalks 100 per cent, developed positive Red Streak infection, the most vigorous infection being obtained where the base of the young leaves of the heart had been inoculated with the suspension and the least vigorous where Red Streaks themselves had been used as inoculum, which possibly may be explained by the comparatively dry nature of the latter.

SERIES IV.

As it was desirable to find out what effect the bacteria would have on the immature tissues of the cane top, a hypodermic syringe fitted with new needle and washers was employed in this series of inoculations. Previous to using for this purpose the syringe had been employed by a dentist for cocaine injections. The syringe was now washed out with methylated spirit and then with distilled water, this being again done after this series of inoculations was completed.

23rd December.—Fresh suspensions of bacteria were made up from seven days old cultures, which were now old and evil-smelling.

Charges of from 5 c.c. to 15 c.c. of suspension were forced into various parts of the cane tops, from the uppermost ligule down to the semi-mature cane stem.

This series of inoculations produced no effect whatever on leaves, heart, leaf-sheaths, or stem. It was therefore concluded that either the syringe still contained fragments of some powerful sterilising agent after washing, or the cultures had become sterile through the action of their own excretions, which is the more likely explanation since the syringe was so often emptied and refilled. Microscopic examination of the suspension used had, through an oversight, been omitted.

Reproduction of Top Rot Symptoms by Artificial Inoculation.

SERIES V.

29th December.—As soon as it was seen that Series IV. had failed to produce any infections, fresh potato-slice cultures were prepared from a vigorous young streak taken from an infected stem in Series I., one control slice being also prepared. Whereas the latter remained free from bacterial growth throughout the period of observation, the former already showed slight viscid, creamy yellow growth after ten hours, and in thirty-six hours sufficient bacterial matter was available for further inoculations; a suspension of this matter was accordingly made in sterilised water, and was found to contain many actively motile bacteria similar in appearance to those previously observed.

30th December.—Twelve stalks in the field mentioned under Series I. were each injected with 1.5 c.c. of the fresh suspension. These inoculations were divided into three groups as follows:—(a) Suspension injected 1 in. below uppermost ligule; (b) suspension injected 3 in. below uppermost ligule; (c) suspension injected 6 in. below uppermost ligule. This grouping really includes quite a variety of inoculation points since some stems were unavoidably further advanced than others.



PLATE 123 (Fig. 6).

The leaves of ten other stalks were inoculated as in Series I. In four of these inoculations the suspension was first passed through the hypodermic syringe. The leaves of six of the stalks in this group were punctured at time of inoculation, while the remainder were inoculated without wounding.

6th January-

Group (a). All showed definite Red Streak infection, many young streaks developing. In one case long light pink and watery marks showed that the infection was descending one young internal white leaf and approaching the growing point of the cane stem.

Groups (b) and (c). - In these stalks all of the internal and some of the external symptoms of Top Rot were produced. The actual symptoms produced in any individual stalk appeared to depend upon the point of injection. Thus where the heart had been inoculated near the growing point of stem the central heart leaves had rotted through; where the growing point itself had received the injection, or part of it, the usual deformities of nodes and internodes (produced since time of inoculation), the reddening of vascular bundles, and reddening and decay of leaf-sheath bases and adjoining stem tissues attendant on a severe attack of Top Rot, were produced; in all cases, regardless of where the suspension had been injected, a most virulent infection which gave off a typical Top Rot odour resulted, and one or more other minor Top Rot symptoms appeared. Unfortunately, the writer was not able to continue his inquiries further, but since the infections all appeared to be so vigorous on this last day of examination it seemed most probable that death of the heart and growing point, and possibly of the stem itself, would have been the final result of the inoculations. No Red Streaks appeared on the leaves of any of the stalks of these two groups. symptoms of Top Rot were produced. The actual symptoms produced in of these two groups.

Summary of Inoculation Results.

The inoculation experiments described above have shown-

- 1. That the formation of Red Streaks can be induced in cane leaves by pricking or scratching the epidermis of either side of the leaf near the base of the blade with a needle which has been drawn through an active Red Streak on a naturally infected leaf.
- 2. That the formation of Red Streaks can be induced in cane leaves by inoculation of the leaves at or near their base with a watery suspension of bacteria taken from a potato-slice culture of the bacteria found in the natural Red Streaks of the field.
- 3. That the formation of Red Streaks can again be induced by the inoculation of cane leaves with a watery suspension of bacteria taken from a potato-slice culture of the bacteria found in Red Streaks already induced by inoculation with the suspension mentioned under 2.
- 4. That the formation of Red Streaks can be induced by placing at the base of the young cane leaves a little of either of the suspensions mentioned under 2 and 3 without bruising or in any way wounding these leaves.
- 5. That the formation of all of the symptoms of Top Rot can be induced by inoculation on the immature tissues of cane stem and leaves with a watery suspension of bacteria taken from a potato-slice culture of the bacteria found in Red Streaks induced by inoculation of leaf with suspension mentioned under 2.

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ILLUSTRATIONS.

- A young stem with heart killed by Top Rot and leaves showing many Red Streaks.
 Portion of a matured leaf about 10 in. above leaf sheath, showing Red Streaks which have developed well up on leaf blade and on midrib.
- 3. A typical Red Streak infected stem.
- 4. A glimpse into a badly Red Streak infected field.
- 5. Stem No. 20 of Series I, of inoculation, showing Red Streaks at base of young leaves.
- A closer view of stem No. 20, with one leaf removed to show Red Streaks. A young watery brown streak is seen forming on leaf marked X.